

# Rac1 Cas9-CKO Strategy

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**Design Date:** 2019-7-24

## **Project Overview**



**Project Name** 

Rac1

**Project type** 

Cas9-CKO

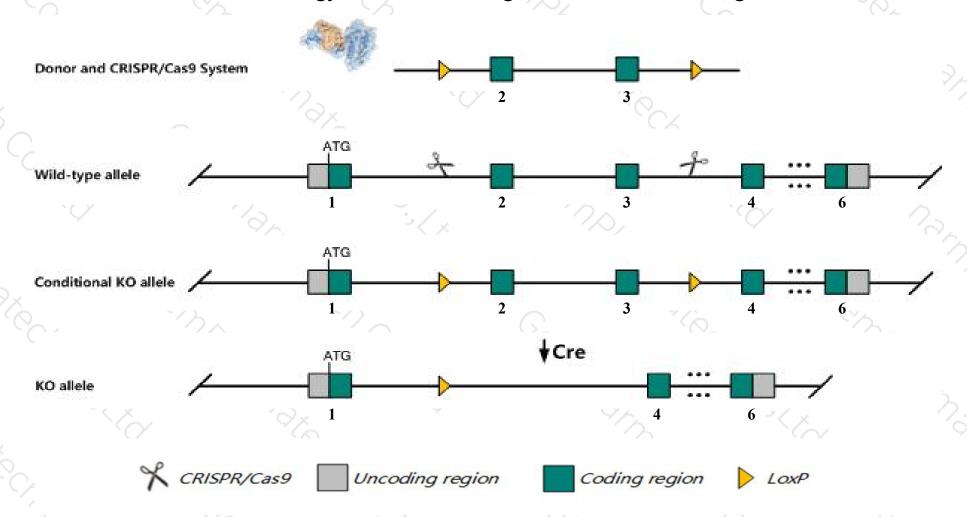
Strain background

C57BL/6JGpt

## Conditional Knockout strategy



This model will use CRISPR/Cas9 technology to edit the *Rac1* gene. The schematic diagram is as follows:



### Technical routes



- ➤ The *Rac1* gene has 4 transcripts. According to the structure of *Rac1* gene, exon2-exon3 of *Rac1-201* (ENSMUST00000080537.13) transcript is recommended as the knockout region. The region contains 190bp coding sequence. Knock out the region will result in disruption of protein function.
- In this project we use CRISPR/Cas9 technology to modify *Rac1* gene. The brief process is as follows:CRISPR/Cas9 system and Donor were microinjected into the fertilized eggs of C57BL/6JGpt mice. Fertilized eggs were transplanted to obtain positive F0 mice which were confirmed by PCR and sequencing. A stable F1 generation mouse model was obtained by mating positive F0 generation mice with C57BL/6JGpt mice.
- The flox mice will be knocked out after mating with mice expressing Cre recombinase, resulting in the loss of function of the target gene in specific tissues and cell types.

### **Notice**



- ➤ According to the existing MGI data, Homozygotes for targeted null mutations exhibit embryonic lethality prior to embryonic day 9.5 with defects in gastrulation. Neutrophil specific knockout mice show defects in inflammatory recruitment and chemotactic responses.
- > The *Rac1* gene is located on the Chr5. If the knockout mice are crossed with other mice strains to obtain double gene positive homozygous mouse offspring, please avoid the two genes on the same chromosome.
- This Strategy is designed based on genetic information in existing databases. Due to the complexity of biological processes, all risk of loxp insertion on gene transcription, RNA splicing and protein translation cannot be predicted at existing technological level.

## Gene information (NCBI)



#### Rac1 Rac family small GTPase 1 [Mus musculus (house mouse)]

Gene ID: 19353, updated on 7-Apr-2019

#### Summary

☆ ?

Official Symbol Rac1 provided by MGI

Official Full Name Rac family small GTPase 1 provided by MGI

Primary source MGI:MGI:97845

See related Ensembl:ENSMUSG00000001847

Gene type protein coding
RefSeq status VALIDATED
Organism Mus musculus

Lineage Eukaryota; Metazoa; Chordata; Craniata; Vertebrata; Euteleostomi; Mammalia; Eutheria; Euarchontoglires; Glires; Rodentia; Myomorpha;

Muroidea; Muridae; Murinae; Mus; Mus

Also known as AL023026, D5Ertd559e

Expression Ubiquitous expression in colon adult (RPKM 163.9), small intestine adult (RPKM 137.6) and 28 other tissuesSee more

Orthologs human all

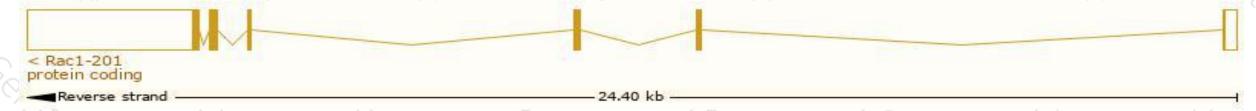
## Transcript information (Ensembl)



The gene has 4 transcripts, all transcripts are shown below:

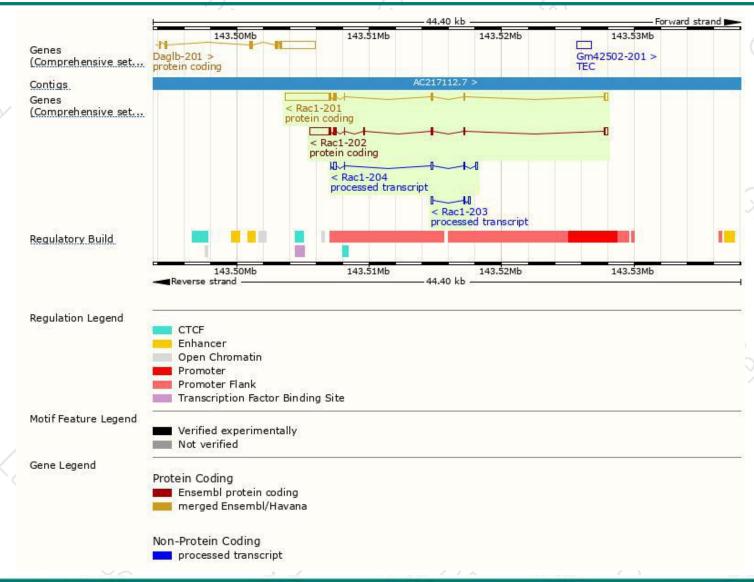
Transcript ID	bp	Protein	Biotype	CCDS	UniProt	Flags
ENSMUST00000080537.13	4159	<u>192aa</u>	Protein coding	CCDS19843	K7Q7T7 P63001	TSL:1 GENCODE basic APPRIS P3
ENSMUST00000100489.3	2325	<u>211aa</u>	Protein coding	CCDS84993	Q3TLP8	TSL:1 GENCODE basic APPRIS ALT1
ENSMUST00000145709.7	585	No protein	Processed transcript	84	-	TSL:3
ENSMUST00000131513.1	344	No protein	Processed transcript	62	22	TSL:5
	ENSMUST000000100489.3 ENSMUST00000145709.7	ENSMUST00000080537.13 4159 ENSMUST00000100489.3 2325 ENSMUST00000145709.7 585	ENSMUST000000100489.3 2325 211aa ENSMUST00000145709.7 585 No protein	ENSMUST00000080537.13         4159         192aa         Protein coding           ENSMUST00000100489.3         2325         211aa         Protein coding           ENSMUST00000145709.7         585         No protein         Processed transcript	ENSMUST00000080537.13         4159         192aa         Protein coding         CCDS19843           ENSMUST00000100489.3         2325         211aa         Protein coding         CCDS84993           ENSMUST00000145709.7         585         No protein         Processed transcript         -	ENSMUST00000080537.13         4159         192aa         Protein coding         CCDS19843         K7Q7T7 P63001           ENSMUST00000100489.3         2325         211aa         Protein coding         CCDS84993         Q3TLP8           ENSMUST00000145709.7         585         No protein         Processed transcript         -         -

The strategy is based on the design of *Rac1-201* transcript, The transcription is shown below



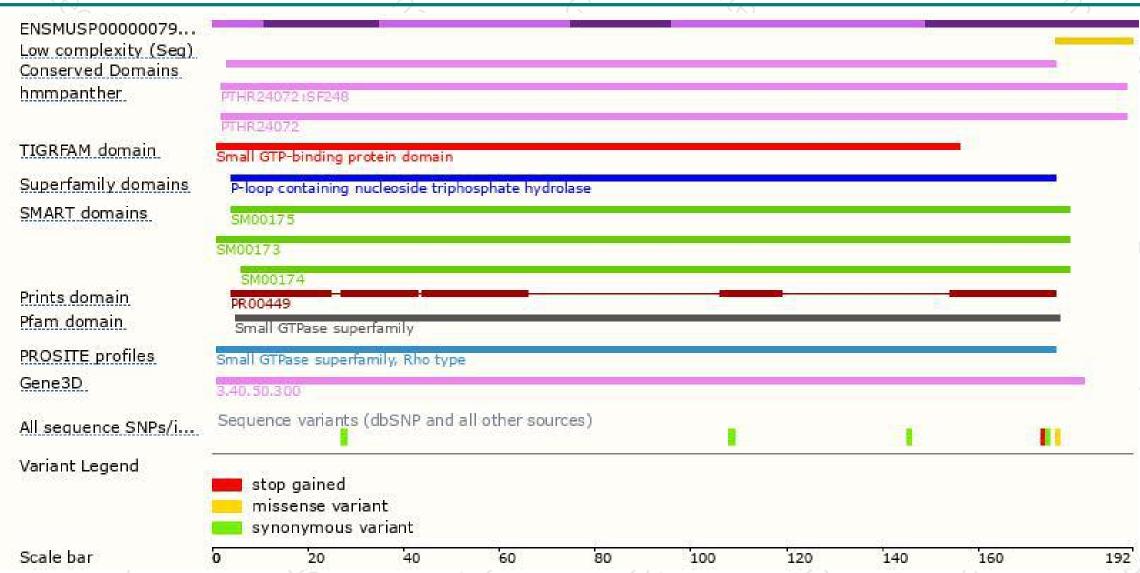
### Genomic location distribution





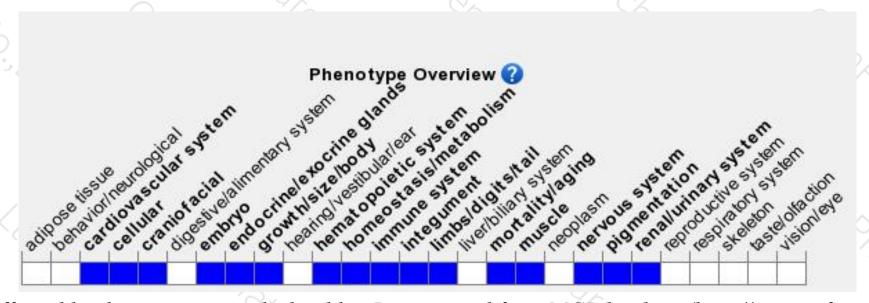
### Protein domain





## Mouse phenotype description(MGI)





Phenotypes affected by the gene are marked in blue.Data quoted from MGI database(http://www.informatics.jax.org/).

According to the existing MGI data, Homozygotes for targeted null mutations exhibit embryonic lethality prior to embryonic day 9.5 with defects in gastrulation. Neutrophil specific knockout mice show defects in inflammatory recruitment chemotactic responses.



If you have any questions, you are welcome to inquire. Tel: 400-9660890





